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Association Between Rotarod Tests and Myelin Basic Protein in the Confirmation of the Cuprizone-Induced Rat Multiple Sclerosis Model and the Relationship of These Parameters with Dietary Zinc Status

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ABSTRACT

Introduction: Experimental autoimmune encephalomyelitis (EAE) is widely used for studies of human inflammatory demyelinating diseases. The cuprizone model is one of the most frequently used. The cuprizone model is a toxic demyelination model. The most significant challenge in this experimental model is demonstrating the development of MS in animals with experimental evidence. Two tests stand out in demonstrating the development of disease in cuprizone-induced rat MS models. The first is the Rotarod test. The Rotarod test is an experimental test that has become frequently used in recent years in the field of neuroscience to demonstrate motor function disorders. The second is the determination of MBP levels at the tissue or blood level. The aim of this study is to investigate the role of rotarod tests and myelin basic protein (MBP) analysis in confirming multiple sclerosis (MS) in a cuprizone-induced rat model, and also the relationship of these parameters with dietary zinc status.

Methods: In the study, approved by the ethics committee, forty-six adult male Wistar rats were divided into five groups. Groups 1 and 2 received Carboxy-methyl-cellulose (CMC) solution. Multiple sclerosis was induced in Groups 3, 4, and 5 by daily gavage of cuprizone in CMC solution (1% of feed intake) for 8 weeks. Group 4 received a zinc-deficient diet, while Group 5 received daily intraperitoneal zinc sulfate supplementation. Myelin basic protein gene expression in animals was determined using Real-Time PCR.

Results: Results showed that MS-induced rats in Groups 3 and 4 exhibited significantly shorter rotarod fall times and higher corpus callosum MBP gene expression compared to other groups ($p<0.05$). Notably, zinc supplementation in Group 5 reversed these effects ($p<0.05$).

Conclusion: These findings confirm that 8 weeks of cuprizone administration induces an MS-like condition in rats, and zinc supplementation effectively ameliorates these MS symptoms.

Keywords: Corpus callosum, cuprizone, multiple sclerosis, myelin basic protein, rotarod tests, zinc

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INTRODUCTION

Multiple sclerosis (MS) is an autoimmune disease characterized by immune-mediated attacks on the central nervous system. This inflammatory process leads to progressive neurodegeneration, resulting in clinical symptoms such as motor deficits, visual disturbances, and impairments in sensation or balance (1). Therefore, there is a growing interest in investigating MS and the problems that arise from the disease using experimental models (1). Cuprizone (bis-cyclohexanone oxaldihydrazone) is a copper chelator and causes copper ions in the system to become completely nonfunctional. When administered orally or through the diet, cuprizone specifically targets mature oligodendrocytes within the central nervous system (2). This administration leads to significant demyelination and a reduction in oligodendrocyte populations, particularly within the corpus callosum and various other cortical regions (2). Consequently, the cuprizone model is frequently utilized in experimental multiple sclerosis (MS) research (3).

Highlights

- **Rotarod performance was impaired in MS groups not receiving supplements.**
- **MBP expression in the corpus callosum increased in MS groups without supplementation.**
- **Thus, MS model was confirmed by rotarod test and MBP mRNA analysis.**
- **Zn treatment improved motor function loss and MBP gene expressions in MS.**
- **Zinc may be an important molecule in the treatment of experimental MS.**

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Given that MS predominantly affects young adults, these models are typically established using young adult mice or rats. In this paradigm, the intake of the toxicant cuprizone induces neuropathological findings that mirror the clinical presentation of MS (3), characterized primarily by widespread demyelination (2,3). Following the cessation of cuprizone exposure, spontaneous remyelination occurs. Thus, the cuprizone-induced MS model provides researchers with a critical platform to both induce targeted demyelination and systematically observe subsequent remyelination processes (2–4).

The most important problem in this experimental model is the demonstration of MS formation in animals with experimental evidence.

Two tests stand out in showing that the disease occurs in cuprizone-induced rat MS models. The first of these is the rotarod test. The rotarod test is an experimental test that has been frequently used in the field of neuroscience for approximately fifteen years to demonstrate motor dysfunctions (5). The second is to determine Myelin Basic Protein (MBP) levels at the tissue or blood level (6).

Zinc is closely related to three basic physiological functions in the body: catalytic, structural and regulatory. Therefore, zinc is an indispensable micronutrient for the normal growth of the body and maintenance of physiological functions (7). Zinc deficiency, which is also a critical element in brain functions, is closely associated with neurodegenerative diseases including MS and impaired cognitive performance (7,8). It has been reported that zinc regulation in biological fluids is significantly impaired in MS patients compared to healthy controls (9,10). Therefore, it can be said that there is an increasing interest in the research of the relationship between MS and zinc. It is also increasingly accepted that zinc in the brain plays important roles in synaptic transmission, paralysis, differentiation and proliferation (11). A report highlights the effects of zinc, a critical micronutrient for brain development and neurological diseases, focusing particularly on its still-unexplored role in white matter disorders, oligodendrocyte damage mechanisms, and myelination processes (12). Similarly, another report suggests that oligodendrocytes can be used as a unique model in research on central nervous system zinc homeostasis; furthermore, it is believed that zinc regulation during development has a decisive effect on cell differentiation (13).

This study, based on the information explained above, has two main purposes:

1. The rotarod test and the determination of MBP gene expression levels in the corpus callosum are used to demonstrate experimental MS formation in rats in which MS was induced by applying cuprizone for eight weeks. Performing these two tests together may be encouraging for future studies by researchers using the experimental MS model.

2. To draw attention to the possible relationships between the dietary status of zinc, which is thought to play an important role in the pathogenesis of MS, as well as its role in brain functions, and MS.

METHODS

Ethics

All experimental procedures were approved by the Animal Ethics Committee of Selçuk University Experimental Medicine Research and Application Center (Approval Code: 17; Approval Date: 2021).

Animals

Adult male Wistar rats (n=46) used in the study were obtained from Selçuk University Experimental Medicine Research and Application Center (SUDAM). All experimental procedures were approved by the animal ethics committee of the same center.

Animals were divided into a total of five groups (group 1: n=6, other four groups n=10).

- **Group 1:** Control
- **Group 2:** Sham multiple sclerosis
- **Group 3:** Multiple sclerosis
- **Group 4:** Multiple sclerosis + zinc deficient
- **Group 5:** Multiple sclerosis + zinc supplemented

Group 1, Group 2, and Group 3 animals were given standard rat chow for 8 weeks.

Group 2 animals were also given cuprizone solvent daily by gavage for 8 weeks.

Procedures

Cuprizone was prepared as described below.

Cuprizone was prepared at a rate of 1% of daily feed intake (0.66 g/kg) with 1% concentration of pure water.

In addition, animals in Group 4 were fed a zinc deficient diet (50 µg/kg zinc).

In addition, animals in Group 5 were given intraperitoneal (ip) zinc sulfate supplement (5 mg/kg/day).

Rotarod Test

Rotarod Tests were performed in SUDAM animal laboratory. In this study, the automatically accelerating May rotarod device (Commat ltd., Ankara/Türkiye) was used to evaluate motor coordination and balance. The apparatus consists of a 7 cm diameter, 30 cm high roller, suitably machined to provide grip, and a power source to rotate the roller. Five circular dividers divide the bar into four equally sized compartments (each 7 cm long).

The rats were given familiarization training 3 days before the evaluation day to help them discover and get used to the rotarod device. Each day, they were allowed to remain on the device for a maximum of 300 seconds per trial, starting with 4 rotations per minute (rpm) and accelerating to a maximum of 20 rpm. If they fell prematurely, they were placed back into the device a maximum of 3 times and the acceleration was started at 4 rpm each time. Each animal was allowed to fail 3 times. The practice trials were terminated after those who were successful completed 300 seconds. The rotarod performances of the animals were recorded on the last day of cuprizone application, which coincided with the fourth day after the acclimation trials. Starting from 4 rpm, the maximum rpm value that the animals could reach was recorded regardless of the time.

Obtaining Tissue Samples from Animals

Animals that were monitored daily for 8 weeks and completed the treatments were sacrificed 24 hours after the last day of treatment and corpus callosum tissue samples were obtained. To prevent the animals from suffering, the animals were killed under general anesthesia with a combination of ketamine (75 mg/kg) and xylazine (10 mg/kg) intraperitoneally. After killing, the rats' brains were quickly removed and frozen on dry ice. The frozen brain tissue was placed in Eppendorf tubes and stored at -80°C until the samples were studied.

RNA Isolation

Total RNA and miRNA isolation from corpus callosum tissues were performed according to commercial kit protocols (Bio Basic, Canada;

miRNAExtractor). Tissues were lysed in 1 ml of lysis buffer using a mechanical homogenizer (SONOPULS mini20, BANDELIN, Germany); purified by spin column technology following chloroform phase separation and ethanol precipitation steps. Quantitative and purity analyses of the obtained RNA samples were performed spectrophotometrically (SMA 1000, Merinton, China); high-quality samples with an OD 260/280 ratio of 1.9 and above were stored at -20°C for further processing.

cDNA Synthesis

cDNA was obtained from total RNA samples for mRNA expression analysis using the iScript cDNA synthesis kit (Bio-Rad, USA) in a thermal cycling machine (Bio-Rad, USA) according to the manufacturer's instructions (25°C for 5 min, 46°C for 20 min, and 95°C for 1 min). After purity checks of the synthesized cDNA, it was stored at -20°C until Real-Time PCR analysis.

Real-Time PCR (qPCR) Analysis

MBP mRNA levels were determined using specific primers (Oligomer, Türkiye). Before quantification, standard curves were created using serial dilutions (between 1/1 and 1/32) prepared from control group samples, and the primer binding temperatures were optimized to ensure standardization of the method. Approximately 50 ng of cDNA was used for each sample in the reactions.

Myelin basic protein gene expression was determined by Real Time PCR method in Selçuk University Faculty of Medicine Molecular Physiology Laboratory (Table 1). qPCR analysis was performed using the 2-ΔΔCT method developed by Livak and Schmittgen (14).

Statistical Methods

Computer package program (IBM Statistical Package for Social Sciences (SPSS) program version 26.0) was used for both Rotarod test and statistical interpretation of MBP gene expression levels. Arithmetic means and standard deviations of the data obtained in the study were calculated. "Shapiro-Wilk" test was applied for homogeneity of data. "One-way analysis of variance (ANOVA)" test was used to determine the differences between the groups. "Duncan" test was used to determine the group from which the difference originated. Differences at the level of P <0.05 were accepted as significant.

Table 1. Primers used for Real Time PCR

Gen	Primer sequence (5'-3')	Function
MBP forward	CTTCAAAGACAGGCCCTCAG	Target gene
MBP back	CCTGTCACCGCTAAAGAAGC	Target gene

MBP: myelin basic protein.

RESULTS

The rotarod test is a standard sensorimotor test used to investigate the motor coordination and learning abilities of animals by measuring the ability of mice to stay on an accelerated rod and run. When the animals were evaluated in terms of the time they stayed on the rod in our study; Animals in the MS group (Group 3) and the zinc-deficient MS group (Group 4) fell off the stick in a shorter time compared to the other groups (p<0.05). Animals in the MS group (Group 5) receiving zinc supplementation remained on the rod longer than animals in Groups 3 and 4 with induced MS (p<0.05). Animals in the control groups (Groups 1 and 2) remained on the rod longer than animals in all other groups (p<0.05; Table 2, Fig. 1).

In an experimental MS model, cuprizone specifically targets oligodendrocytes and causes demyelination in the corpus callosum. MBP, abundant in the outermost layer of the myelin sheath surfaces of oligodendrocytes, has been suggested as one of the critical autoantigens that can support immune responses and may also be associated with autoimmune responses.

In our study, we obtained the highest MBP gene expression levels in the corpus callosum in Group 4 animals fed with MS-induced zinc-deficient diet and Group 3 animals fed with MS-induced standard rat chow (p<0.05). Zinc supplementation significantly reduced myelin basic protein gene expression levels in Group 5 animals with MS-induced compared to Groups 3 and 4, bringing them to control values (Groups 1 and 2) (p<0.05; Table 2, Fig. 2).

DISCUSSION

Discussing Rotarod Performance Test Findings

The rotarod test is a standard sensorimotor test used to investigate the motor coordination and learning abilities of animals by measuring the ability of mice to stay on an accelerated rod and run (15). The rotarod test was first used to measure motor coordination in rodents in 1957 (16). These tests were later used to measure both motor performance and learning skills in rat experimental models treated with various substances (17). Rotarod test has become one of the most frequently used motor and cognitive function tests today (15,17).

Table 2. Rotarod test and corpus callosum myelin basic protein (MBP) gene expression levels of the study groups

Groups	ROTAROD TEST Remaining time on the stick "seconds" Mean ± SD	MBP ($2^{-\Delta\Delta CT}$) Mean ± SD
Control group (G1)	209.66±50.06**a**	0.95±0.13b
Sham group (G2)	178.50±87.44**a**	0.98±0.09b
MS group (G3)	58.60±23.66**c**	1.58±0.13a
Zn deficient MS group (G4)	35.00±16.61**c**	1.59±0.09a
Zinc supplemented MS group (G5)	89.40±50.83**b**	1.09±0.16b

a >b >c: The difference between averages with different letters in the same column is important (P <0.05). MS: multiple sclerosis; MBP: myelin basic protein; Zn: zinc.
P-Values, ROTAROD; G1-G2=1.000, G1-G3=0.795, G1-G4=0.018, G1-G5=0.990, G2-G3=0.769, G2-G4=0.047, G2-G5=0.962, G3-G4=0.755, G3-G5=0.366, G4-G5=0.032
P-Values MBP: G1-G2=0.990 G1-G3=0.000, G1-G4=0.000, G1-G5=0.260, G2-G3=0.000, G2-G4=0.000, G2-G5=0.368, G3-G4=1.000 G3-G5=0.000, G4-G5=0.000

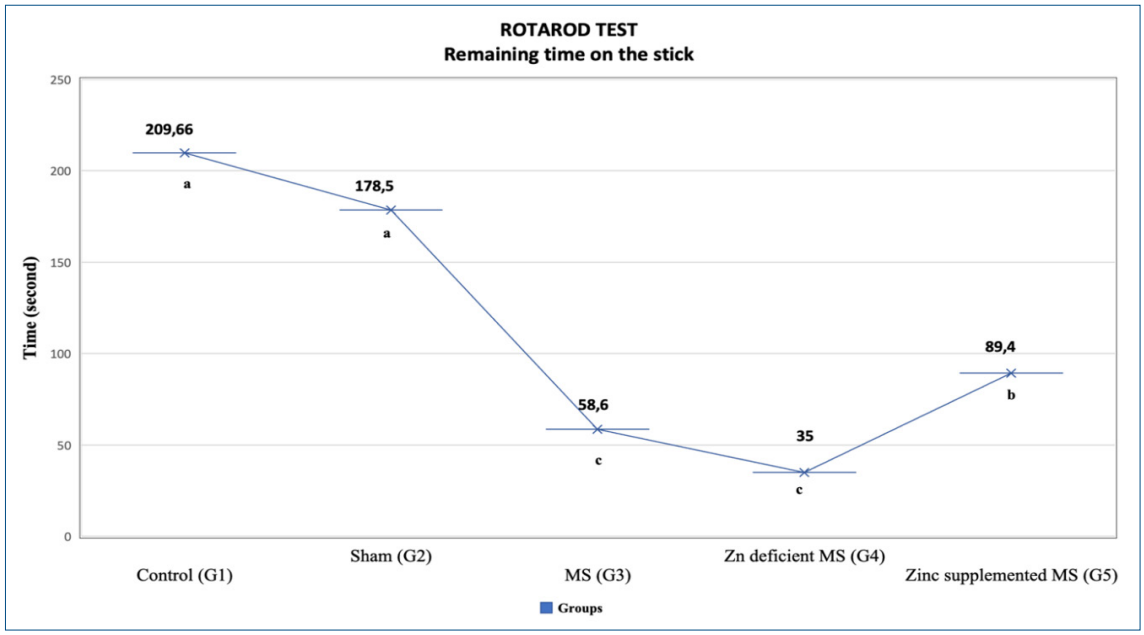

Figure 1. Rotarod test results of the study groups. a >b >c: means with different letters are statistically significant. (P <0.05).

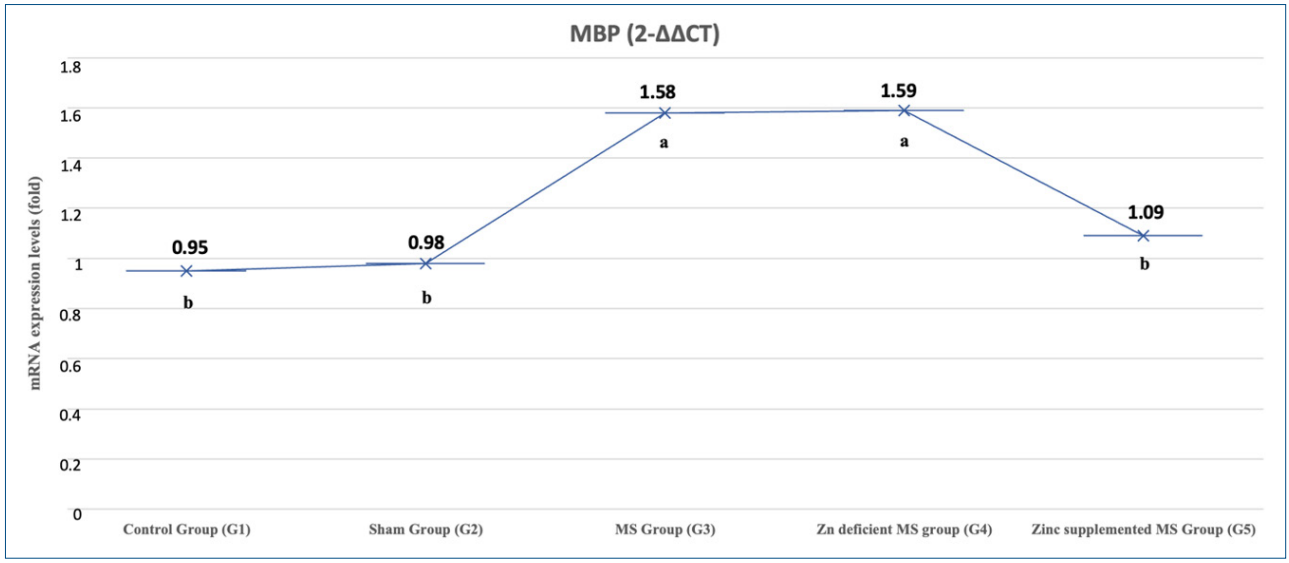

Figure 2. Myelin basic protein (MBP) gene expression levels in the Corpus Callosum Tissue of the study groups. a >b: Means with different letters are statistically significant. (p<0.05).

In our study, when the animals were compared in terms of Rotarod tests (time spent on the rod); the animals in the MS group (Group 3) and the MS group (Group 4) fed with a zinc deficient diet fell off the rod in a shorter time than the other groups. This finding shows that Group 3 and Group 4 animals, which were created as experimental MS models, had weaker motor coordination than Group 5 and control group animals receiving zinc supplements. In our study, although the rotarod performance test of the animals in the zinc-deficient diet-fed MS group (Group 4) was numerically lower than the cuprizone-induced MS group (Group 3), it was not statistically significant. This seems likely to be due to individual differences in the animals' rotarod tests.

The rotarod test is one of the most preferred tests to demonstrate motor dysfunction in cuprizone-induced rat MS models (17). Han et al. (18) applied the rotarod test to determine the loss of motor function in the cuprizone-induced demyelination mouse model and showed that cuprizone animals that did not receive treatment fell on the rod in a short time and exhibited impairment in motor functions. In a study investigating the effects of enriched environment on MS in the cuprizone-induced mouse MS model, it was shown that the rotarod performance of animals not exposed to enriched environment was poorer, that is, they fell on the rod in a shorter time (19). As a result, rotarod performance is applied as a reliable test in determining motor functions in experimental MS models (20,21). In our study, it was observed that the rotarod performances of the MS groups that did not receive zinc supplementation (Groups 3 and 4) decreased significantly compared to the other groups, indicating deterioration in motor functions. This finding we obtained in the current study is important evidence that cuprizone application causes MS in rats.

The most important finding regarding this parameter in our study is that the rotarod performance of Group 5 MS animals receiving zinc supplementation is better than the MS groups not receiving

supplementation (Groups 3 and 4). This finding we obtained regarding zinc supplementation shows that zinc can prevent the deterioration in motor functions that occurs in MS disease. In our Med-Line searches, we could not find any study investigating how dietary zinc status affects motor functions in MS disease. Our finding showing that zinc supplementation can prevent the deterioration in motor functions in the cuprizone-induced rat MS model may be a guide for future studies investigating the relationship between MS and zinc.

Discussion of MBP Gene Expression Levels in Corpus Callosum Tissue

Multiple sclerosis is an autoimmune central nervous system disease characterized by inflammation, demyelination and axonal damage. In the case of MS, the immune system attacks the myelin sheath that protects the nerve fiber, leading to demyelination (22). Myelin basic protein, which is abundant in the outermost layer of the myelin sheath surfaces of oligodendrocytes, has been suggested as one of the critical autoantigens that can both support the immune response and may be associated with autoimmune events (23). High MBP expression was described in MS patients during disease activity (23). It was therefore noted that the increase in MBP expression could be a reliable marker in MS disease (24). In the current study, the MBP gene expression levels in the corpus callosum of the MS groups without zinc supplementation (Groups 3 and 4) were significantly higher than all other groups. In their study, Tian et al. (6) showed that both CSF and serum MBP levels increased significantly in MS patients compared to controls. The same researchers also suggested in their aforementioned studies that evaluation of CSF and serum MBP levels may enable the accurate diagnosis of MS disease. Therefore, they suggested that determination of both CSF and serum MBP levels may be a critical marker for the diagnosis of MS (6). Similarly, Agliardi et al. (25), considering that MBP levels are high in MS patients, reported that a minimally invasive blood test measuring MBP concentration could be a promising tool that could facilitate the diagnosis of MS (25).

In our study, increased MBP gene expression levels in the corpus callosum in MS groups that did not receive zinc supplementation (Groups 3 and 4) can be considered as evidence that cuprizone application causes MS.

The most critical point to emphasize in our study is that MBP gene expression levels in the MS group receiving zinc supplementation (Group 5) were lower than in the MS groups not receiving supplementation (Groups 3 and 4). Again, the MBP gene expression levels of the MS group (Group 5) receiving zinc supplementation were not different from Group 1 (control) and Group 2 (Sham). This finding we obtained is a very critical finding. Because there is no study investigating how dietary zinc status affects MBP gene expression levels in the corpus callosum in MS disease. Given that zinc plays a critical role in motor functions as well as in important events such as synaptic transmission and suppression of neuroinflammation, our finding is a crucial result in the zinc-MS relationship (26). The current study is the first to reveal the relationship of dietary zinc status with MBP gene expression levels in MS disease. In our study, we determined MBP gene expression in the corpus callosum only as a marker of the experimental MS model. We also considered the decrease in MBP gene expression in the corpus callosum in MS animals receiving zinc supplementation as a result of zinc's regulatory rather than suppressive effect.

When the results of our study are evaluated as a whole: Impaired Rotarod tests and increased MBP gene expression in the corpus callosum indicate that cuprizone administration causes experimental MS. Zinc supplementation reversed the above-mentioned symptoms in the MS rat model.

As a result, in our study rotarod test and MBP gene expression analysis in the corpus callosum were applied to determine whether the experimental MS model was realized in rats. While the rotarod performances of MS groups (Groups 3 and 4) that did not receive zinc supplementation were significantly suppressed; Again, MBP gene expressions in the corpus callosum of MS groups that did not receive supplements (Groups 3 and 4) were found to be higher than all other groups.

These findings show that the cuprizone-induced MS model was successfully implemented in our study.

Zinc supplementation (Group 5) improved motor function loss compared to non-supplemented groups (Groups 3 and 4).

The increased MBP gene expressions in the corpus callosum in the groups not receiving supplementation (Groups 3 and 4) were reached to control values with zinc supplementation (Group 5).

As a result of our study, the loss of motor function observed in the MS groups that did not receive supplementation (Groups 3 and 4) and the changes in increased MBP gene expression in the corpus callosum were eliminated by zinc supplementation.

The findings of this study indicate that zinc may be an important molecule in experimental MS models.

Limitations

In this study, confirmation of demyelination in corpus callosum tissue using methods such as Luxol Fast Blue could not be achieved. Furthermore, immunohistochemical staining of MBP and oligodendrocytes such as Olig2 could not be performed. Determining these parameters in future studies may provide us with more original information.

Ethics Committee Approval: Experimental Animals Ethics Committee of Selçuk University Experimental Medicine Research and Application Center (Approval Code: 17, Approval Date: 2021)

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Conflict of Interest: The authors declared that there is no conflict of interest.

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REFERENCES

1. Shahidi SH, Kordi MR, Rajabi H, Malm C, Shah F, Quchan ASK. Exercise modulates the levels of growth inhibitor genes before and after multiple sclerosis. J Neuroimmunol. 2020;341:577172. [Crossref]
2. Abdel-Maged AE, Gad AM, Rashed LA, Azab SS, Mohamed EA, Awad AS. Repurposing of secukinumab as neuroprotective in cuprizone-induced multiple sclerosis experimental model via inhibition of oxidative, inflammatory, and neurodegenerative signaling. Mol Neurobiol. 2020;57:3291–3306. [Crossref]
3. Basoglu H, Boylu NT, Kose H. Cuprizone-induced demyelination in Wistar rats; electrophysiological and histological assessment. Eur Rev Med Pharmacol Sci. 2013;17:2711–2717.
4. Martinez B, Peplow PV. Protective effects of pharmacological therapies in animal models of multiple sclerosis: a review of studies 2014-2019. Neural Regen Res. 2020;15:1220–1234. [Crossref]
5. van den Berg R, Laman JD, van Meurs M, Hintzen RQ, Hoogenraad CC. Rotarod motor performance and advanced spinal cord lesion image analysis refine assessment of neurodegeneration in experimental autoimmune encephalomyelitis. J Neurosci Methods. 2016;262:66–76. [Crossref]
6. Tian ZJ, Zhao XX, Li ZH, Zhang F, Cao FT, Li SM, et al. Evaluation of myelin basic protein levels with receiver operating characteristic curves for diagnosis of multiple sclerosis. Nan Fang Yi Ke Da Xue Xue Bao. 2009;29:250–252.
7. Bredholt M, Frederiksen JL. Zinc in multiple sclerosis: a systematic review and meta-analysis. ASN Neuro. 2016;8. [Crossref]

8. Elitt CM, Fahrni CJ, Rosenberg PA. Zinc homeostasis and zinc signaling in white matter development and injury. Neurosci Lett. 2019;707:134247. [Crossref]
9. Choi BY, Jung JW, Suh SW. The emerging role of zinc in the pathogenesis of multiple sclerosis. Int J Mol Sci. 2017;18. [Crossref]
10. Nirooei E, Kashani SMA, Owrangi S, Malekpour F, Niknam M, Moazzen F, et al. Blood trace element status in multiple sclerosis: a systematic review and meta-analysis. Biol Trace Elem Res. 2022;200:13–26. [Crossref]
11. Choi BY, Kim IY, Kim JH, Kho AR, Lee SH, Lee BE, et al. Zinc transporter 3(ZnT3) gene deletion reduces spinal cord white matter damage and motor deficits in a murine MOG-induced multiple sclerosis model. Neurobiol Dis. 2016;94:205–212. [Crossref]
12. Liu S, Wang N, Long Y, Wu Z, Zhou S. Zinc homeostasis: an emerging therapeutic target for neuroinflammation related diseases. Biomolecules. 2023;13:416. [Crossref]
13. Elitt CM., Ross MM, Wang J, Fahrni CJ, Rosenberg PA. Developmental regulation of zinc homeostasis in differentiating oligodendrocytes. Neurosci Lett. 2024;831;137727. [Crossref]
14. Livak KJ, Schmittgen TD. Analysis of relative gene expression data using real-time quantitative PCR and the 2(-Delta Delta C (T)) method. Methods. 2001;25:402–408. [Crossref]
15. Tashakori A, Hassanpour S, Vazir B. Protective effect of crocin on cuprizone-induced model of multiple sclerosis in mice. Naunyn Schmiedebergs Arch Pharmacol. 2023;396:1713–1725. [Crossref]
16. Dunham NW, Miya TS. A note on a simple apparatus for detecting neurological deficit in rats and mice. J Am Pharm Assoc Am Pharm Assoc. 1957;46:208–209. [Crossref]
17. Lubrich C, Giesler P, Kipp M. Motor behavioral deficits in the cuprizone model: validity of the Rotarod Test Paradigm. Int J Mol Sci. 2022;23. [Crossref]
18. Han SR, Kang YH, Jeon H, Lee S, Park SJ, Song DY, et al. Differential expression of miRNAs and behavioral change in the cuprizone-induced demyelination mouse model. Int J Mol Sci. 2020;21. [Crossref]
19. Mohamed A, Al-Kafaji G, Almahroos A, Almosawi Z, Alalwan H, Abdulla R, et al. Effects of enhanced environment and induced depression on cuprizone mouse model of demyelination. Exp Ther Med. 2019;18:566–572. [Crossref]
20. Franco-Pons N, Torrente M, Colomina MT, Vilella E. Behavioral deficits in the cuprizone-induced murine model of demyelination/remyelination. Toxicol Lett. 2007;169:205–213. [Crossref]
21. Moore SM, Khalaj AJ, Kumar S, Winchester Z, Yoon J, Yoo T, et al. Multiple functional therapeutic effects of the estrogen receptor beta agonist indazole-CI in a mouse model of multiple sclerosis. Proc Natl Acad Sci U S A. 2014;111:18061–18066. [Crossref]
22. Ganesan A, Muralidharan P, Ramya LN. The fulcrum of demyelination in multiple sclerosis. Curr Protein Pept Sci. 2023;24:579–588. [Crossref]
23. Gutierrez-Fernandez M, de la Cuesta F, Tallon A, Cuesta I, Fernandez-Fournier M, Laso-Garcia F, et al. Potential roles of extracellular vesicles as biomarkers and a novel treatment approach in multiple sclerosis. Int J Mol Sci. 2021;22. [Crossref]
24. Torres Iglesias G, Fernandez-Fournier M, Botella L, Piniella D, Laso-Garcia F, Carmen Gomez-de Frutos M, et al. Brain and immune system-derived extracellular vesicles mediate regulation of complement system, extracellular matrix remodeling, brain repair and antigen tolerance in multiple sclerosis. Brain Behav Immun. 2023;113:44–55. [Crossref]
25. Agliardi C, Guerini FR, Zanzottera M, Bolognesi E, Picciolini S, Caputo D, et al. Myelin basic protein in oligodendrocyte-derived extracellular vesicles as a diagnostic and prognostic biomarker in multiple sclerosis: a pilot study. Int J Mol Sci. 2023;24. [Crossref]
26. Sun R, Wang J, Feng J, Cao B. Zinc in Cognitive impairment and aging. Biomolecules. 2022;12:1000. [Crossref]